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Kenneth Solomon
Type or Print Name


Signature

In re application of: Rees :
U.S. National Stage for: PCT/GB01/00241 :
Filed: January 22, 2001 :
For: PHYSIOLOGICAL MEDIUM FOR :
PERFUSING, PRESERVING AND STORING :
ISOLATED CELL, TISSUE AND ORGAN :
SAMPLES :

BOX PCT
Assistant Commissioner for Patents
Washington, D.C. 20231

PRELIMINARY AMENDMENT

This is filed as a Preliminary Amendment concurrently with the above-referenced national stage filing of PCT/GB01/00241. It is respectfully requested that the amendments to the claims as described below be entered into the case and that the amended claims be examined on the merits.

In the Claims

The claims of the application have been amended herein as indicated in the following marked up copies of the claims wherein the "[]" symbols in line 2 of Claims 9 and 11 are part of the claim text and not intended to indicate deletions:

3. (Amended) A physiological medium as claimed in Claim 1 [or 2] wherein the salt component comprises:

- (c) from 1.0 to 2.5 mmoles/L of calcium ions, and
 - (d) from 0.4 to 2.4 mmoles/L of magnesium ions.

4. (Amended) A physiological medium as claimed in [any of claims] claim 1 [to 3] wherein the salt component comprises

135.32 mmoles /L of sodium ions, 5.00 mmoles /L of potassium ions, 1.25 mmoles /L of calcium ions, 0.45 mmoles /L of magnesium ions, as chloride salts, and 118.40 mmoles /L of chloride ions as sodium, potassium, calcium and magnesium salts.

5. (Amended) A physiological medium as claimed in [any one of Claims] claim 1 [to 4] wherein the buffer component comprises 25.00 mmoles /L of bicarbonate ions as sodium salt and 5.0 mmoles /L of N,N-bis (2-hydroxy ethyl)-2-amino-ethanesulfonic acid (BES).

6. (Amended) A physiological medium as claimed in [any of Claims] claim 1 [to 5] wherein the substrate component comprises 10 mmoles /L of D-glucose, 110 μ moles /L of glycerol and 10.0 μ moles /L of choline as the chloride salt.

7. (Amended) A physiological medium claimed in [any of Claims] claim 1 [to 6] wherein the amino acid component comprises 300 μ moles /L of L-glutamate as sodium salt, 20 μ moles /L of L-aspartate as sodium salt and 400 μ moles /L of L-glutamine.

8. (Amended) A physiological medium claimed in [any of Claims] claim 1 [to 7] wherein the co-enzyme component comprises 40.0 nmoles /L of thiamine as thiamine pyrophosphate chloride.

9. (Amended) A physiological medium as claimed in [any one of Claims] claim 1 [to 8] wherein the vitaminoid component comprises 50.0 gmoles /L of [-]-p-hydroxy- γ -trimethylaminobutyrate hydrochloride (L-carnitine).

10. (Amended) A physiological medium as claimed in [any one of Claims] claim 1 [to 9] wherein the protein component comprises 28.0 m. I.U./L of recombinant human insulin (expressed in E.coli).

11. (Amended) A physiological medium as claimed in [any one of Claims] claim 1 [to 10] wherein the antibiotic component comprises 100 mg/L of D-[-]-theo-2-dicWoroacetamide-I-(p-nitrophenyl)-1,3-propane acid (chloramphenicol).

12. (Amended) A method for producing a physiological medium according to [any one of Claims] claim 1 [to 11] which comprises adding in the following order: sodium chloride, potassium chloride, calcium chloride, magnesium chloride, the TES, MOPS, or BES, thiamine, carnitine, choline, glycerol, insulin, aspartate, glucose, glutamate, glutamine, and sodium bicarbonate to sterile purified water, with constant stirring, making up to the desired volume, filtering and storing in sterile sealed vessels.

13. (Amended) Concentrates for the preparation of a physiological medium as claimed in [any one of Claims] claim 1 [to 11] which comprise the salt, buffer, substrate, amino acid, co-enzyme, vitaminoid and protein components, and dilutable with sterile purified water to form said physiological medium.

14. (Amended) Concentrates for the preparation of a physiological medium as claimed in [any one of Claims] claim 1 [to 11] which comprise the salt, buffer, substrate, amino acid, co-enzyme, vitaminoid and protein components, except for sodium bicarbonate, and dilutable with sterile purified water with the addition of sodium bicarbonate to form said physiological medium.

Remarks

The claims have been amended to bring them into conformity with U.S. practice.
Early and favorable consideration is respectfully requested.

Respectfully submitted,



Kenneth Solomon
Reg. No. 31,427
Howell & Haferkamp, L.C.
7733 Forsyth Boulevard, Suite 1400
St. Louis, Missouri 63105
(314) 727-5188

September 17, 2001

AMENDED CLAIMS

3. (Amended) A physiological medium as claimed in Claim 1 wherein the salt component comprises:

- (c) from 1.0 to 2.5 mmoles/L of calcium ions, and
- (d) from 0.4 to 2.4 mmoles/L of magnesium ions.

4. (Amended) A physiological medium as claimed in claim 1 wherein the salt component comprises

135.32 mmoles /L of sodium ions, 5.00 mmoles /L of potassium ions, 1.25 mmoles /L of calcium ions, 0.45 mmoles /L of magnesium ions, as chloride salts, and 118.40 mmoles /L of chloride ions as sodium, potassium, calcium and magnesium salts.

5. (Amended) A physiological medium as claimed in claim 1 wherein the buffer component comprises 25.00 mmoles /L of bicarbonate ions as sodium salt and 5.0 mmoles /L of N,N-bis (2-hydroxy ethyl)-2-amino-ethanesulfonic acid (BES).

6. (Amended) A physiological medium as claimed in claim 1 wherein the substrate component comprises 10 mmoles /L of D-glucose, 110 μ moles /L of glycerol and 10.0 μ moles /L of choline as the chloride salt.

7. (Amended) A physiological medium claimed in claim 1 wherein the amino acid component comprises 300 μ moles /L of L-glutamate as sodium salt, 20 μ moles /L of L-aspartate as sodium salt and 400 μ moles /L of L-glutamine.

8. (Amended) A physiological medium claimed in claim 1 wherein the co-enzyme component comprises 40.0 nmoles /L of thiamine as thiamine pyrophosphate chloride.

9. (Amended) A physiological medium as claimed in claim 1 wherein the vitaminoid component comprises 50.0 gmoles /L of [-]-p-hydroxy- γ - trimethylaminobutyrate hydrochloride (L-carnitine).

10. (Amended) A physiological medium as claimed in claim 1 wherein the protein component comprises 28.0 m. I.U./L of recombinant human insulin (expressed in E.coli).

11. (Amended) A physiological medium as claimed in claim 1 wherein the antibiotic component comprises 100 mg/L of D-[-]-theo-2-dicloroacetamide-I-(p-nitrophenyl)-1,3-propane acid (chloramphenicol).

12. (Amended) A method for producing a physiological medium according to claim 1 which comprises adding in the following order: sodium chloride, potassium chloride, calcium chloride, magnesium chloride, the TES, MOPS, or BES, thiamine, carnitine, choline, glycerol, insulin, aspartate, glucose, glutamate, glutarnine, and sodium bicarbonate to sterile purified water, with constant stirring, making up to the desired volume, filtering and storing in sterile sealed vessels.

13. (Amended) Concentrates for the preparation of a physiological medium as claimed in claim 1 which comprise the salt, buffer, substrate, amino acid, co-enzyme, vitaminoid and protein components, and dilutable with sterile purified water to form said physiological medium.

14. (Amended) Concentrates for the preparation of a physiological medium as claimed in claim 1 which comprise the salt, buffer, substrate, amino acid, co-enzyme, vitaminoid and protein components, except for sodium bicarbonate, and dilutable with sterile purified water with the addition of sodium bicarbonate to form said physiological medium.